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OBSERVATIONS ON THE MARINE LEECH *CALLIOBDELLA CAROLINENSIS* (HIRUDINEA: PISCICOLIDAE), EPIZOOTIC ON THE ATLANTIC MENHADEN [1]

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Apart from the biological accounts of three European leeches, *Crangonobdella murmanica* by Selensky (1923), *Hemibdella soleae* by Llewellyn (1965) and *Oceanobdella blennii* by Gibson and Tong (1969) and Sawyer (1970), almost nothing is known about the biology of marine leeches, particularly from North America. This lack of information is partially due to their obscure taxonomy and partially due to the difficulty of working with these elusive parasites. Starting in the winter of 1970–71 an epidemic of a new piscicolid, *Calliobdella carolinensis,* on the Atlantic menhaden, *Brevoortia tyrannus,* gave us an unusual opportunity to investigate the behavior and population fluctuations of this marine leech (Sawyer and Chamberlain, 1972). Presented below are the results of three years of observations on various aspects of the biology of *C. carolinensis* in the vicinity of Charleston, South Carolina. The study constitutes the first account of a marine leech epizootic outbreak (epidemic).

METHODS

The majority of the observations was confined to two localities: (1) Beresford Creek, the type locality of *C. carolinensis:* and (2) the south end of Folly Beach. Both localities were chosen because of the accessibility and abundance of the leeches, as well as the prominent ecological differences between them.

Beresford Creek (32°53.2′N; 79°52.7′W) is a short estuarine tributary of the Wando River, Charleston County, South Carolina. The salinity is normally about 12–16‰. Like most estuarine streams in coastal South Carolina, the bottom and shoreline consist mainly of mud, mixed with occasional clumps of oysters. The water itself is heavily silted and contains many planktonic organisms which constitute food for the schools of juvenile menhaden which abound in the creek. The leeches were collected by periodically netting its host, the Atlantic menhaden, in a $20' \times \frac{1}{2}''$ square mesh otter trawl towed for approximately 2 hours. To determine infestation rates, the menhaden were immediately isolated into plastic bags for later laboratory examination. The unattached leeches found in the bottom of the boat and on shells, were collected for behavioral studies. Large samples of other species of fish collected along with the menhaden were also examined for leeches. The otter trawl technique was most successful in the winter months, presumably because the normally fast-swimming menhaden were too lethargic to escape the net.

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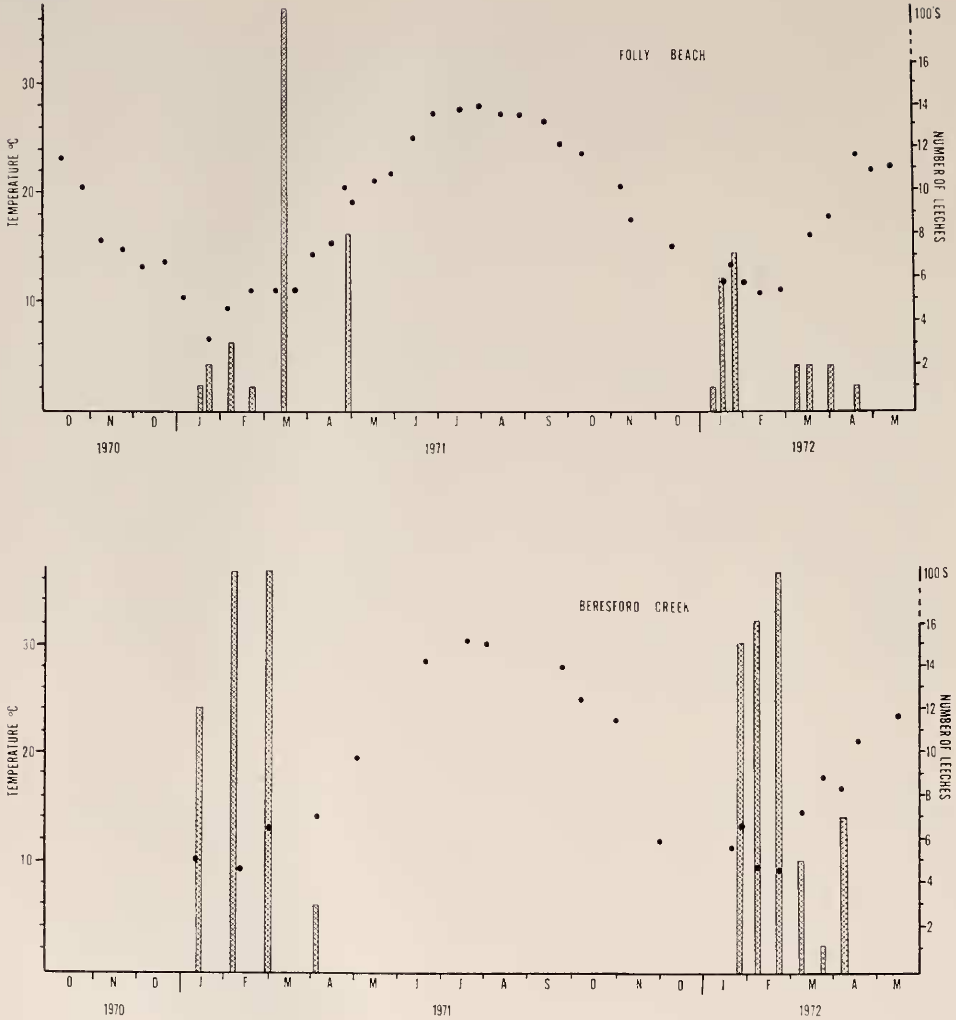

FIGURE 1. Temperature and seasonal occurrence of *Calliobdella carolinensis* at Folly Beach (upper graph) and at Beresford Creek (lower graph) from October 1970 to May 1972. The solid dots indicate the temperature of the water (°C) when each sample was taken. The vertical bars indicate the number of leeches collected on seaweeds at Folly Beach and on menhaden at Beresford Creek.

The south end of Folly Beach (32°38.3′N; 79°58.5′W) constitutes the north bank of the mouth of the estuarine Folly River. The salinity was normally about 28–32‰, but varied from 25.8–35.0‰. The beach area is usually silty and turbulent. In a one hour period the leeches were hand collected on the green seaweeds (primarily *Ulva lactuca*) free-floating in the open surf. During the winter months a few leeches were collected on menhaden caught in a 65′ × 6′ × ¼″ square mesh bag seine dragged in the surf as a part of a separate study on the surf zone fish of Folly Beach.

In addition to the above observations, our understanding of the distribution of this leech is augmented by random collections at various localities along the North and South Carolina coasts either by capturing menhaden in nets or by sampling free-floating green seaweeds. Several attempts to collect the leeches with plankton nets (1 m diameter, 0.5 mm square mesh) were also successful in certain tidal creeks near Charleston.

In the laboratory the leeches were placed into aquaria (salinity 28‰) of various sizes in a refrigerator and maintained in constant dark at 9° C. The leeches, which were not fed, thrived for months under these conditions.

Results

Population dynamics and distribution

In spite of extensive research on the parasites of the Altantic menhaden, *C. carolinensis* was practically unknown until the population explosion of January–March 1971 in which hundreds, if not thousands, of leeches were found with almost every collection of menhaden. Local commercial fisherman verified that during this period the leeches accumulated on the decks of their boats in great numbers and were often found on the hands and feet of the fishermen. Prior to this population explosion the only records of *C. carolinensis* in South Carolina were three individuals collected unattached to a host in Charleston Harbor near Fort Johnson on 14 March 1963, and one individual collected from a menhaden at Dewees Inlet, Isle of Palms, South Carolina on 9 February 1970. The infestations of January–March 1972 and 1973, while still high, had declined noticeably from the same period in 1971.

C. carolinensis displays a remarkable seasonal occurrence which correlates with the temperature of the sea water (Fig. 1). For example, at Beresford Creek during the two full winters of the study, the first individuals of *C. carolinensis* were encountered on menhaden on 12 January 1971 and 26 January 1972 when the water temperature had dropped to 10.1° C and 13.3° C, respectively. On 13–14 December 1972 (water 15.0°, 16.8° C, respectively) small individuals were found on the Blueback herring, *Alosa aestivalis* (see discussion). At Folly Beach the first individuals were encountered, unattached to a host, on 12 January 1971 and 9 January 1972, when the water temperature had dropped to 9.5° C and 14.9° C, respectively.

The presence of the first leeches also strongly correlates with the presence of menhaden in the various collecting nets. At Beresford Creek the first menhaden were collected in the otter trawl on the same dates mentioned above. At Folly Beach the first menhaden were collected on 21 January 1971 and 23 January 1972, when the water temperature was 6.4° C and 13.2° C, respectively. It is known that menhaden occur in these regions throughout the year, but are caught in the nets in great number only from January to early March, presumably because the fish become lethargic in the cold water (see discussion). This lethargy of the menhaden probably plays an important role in the life cycle of *C. carolinensis.*

Leeches reached their greatest abundance when the water was near its coldest temperature. For example, at Beresford Creek during the two full winters of the study the greatest number of individuals were encountered on menhaden on 2 March 1971 and 23 February 1972, when the water was 13.1° C and 9.4° C, respectively. At Folly Beach the greatest number of individuals were encountered on seaweeds on 13 March 1971 and 29 January 1972, when the water temperature was 10.8° C and 11.7° C, respectively.

With the advent of rapidly warming water, the leeches became scarcer and eventually disappeared by the end of April. For example, at Beresford Creek the last individuals were encountered on menhaden on 5 April 1971 and 7 April 1972 when the water was 14.4° C and 17.0° C, respectively. At Folly Beach the last individuals were encountered on seaweeds 29 April 1971 and 19 April 1972, when the water was 17.0° C and 23.2° C, respectively. An attempt on 1 June 1972 to collect menhaden from a commercial shrimp trawler in the open sea three miles off Folly Beach yielded 44 large (137–207 mm, SL) uninfected menhaden. A similar attempt on 29 October yielded 67 uninfected menhaden (128–197 mm, SL). Collections of menhaden throughout the summer and fall of 1972 failed to turn up any leeches. In fact in 1970, 1971 and 1972 no leeches were encountered from May until the following mid-December or early January in spite of repeated attempts to collect them on the seaweeds or menhaden. However, relatively few menhaden were collected in the estuarine creeks during this period possibly because of the avoidance of the otter trawl by the fast-swimming menhaden (Wilkens and Lewis, 1971). Menhaden were collected during this time by local fishermen using other techniques.

The population structures of both the Beresford Creek and the Folly Beach populations of *C. carolinensis* were examined by measuring the total lengths of relaxed individuals with the aid of an ocular micrometer. At Beresford Creek 210 individuals, ranging in length from 3.0 to 29.0 mm (mode, 9.0 mm), were collected on 23 February 1972 from the mouth cavities of 41 menhaden. At Folly Beach 91 unattached individuals, ranging in length from 5.0 to 25.0 mm (modes, 11.0 and 17.5 mm) were collected on 13 March 1971 from a large clump of floating *Ulva*.

The Beresford Creek population, taken directly from the host, contained more immature (3 to 9 mm) individuals than the unattached Folly Beach population. This is consistent with the normal behavior of the Piscicolidae, *i.e.*, the mature individuals are the first to leave the host to breed. Becker and Katz (1965a) made similar observations on *Piscicola salmositica*.

In South Carolina *C. carolinensis* is restricted to the mouths and inlets of estuarine rivers and streams from North Inlet, Georgetown, south to Hunting Island State Park below Beaufort. It has not yet been found south of South Carolina, nor does it occur along the lengthy, sandy beaches of the Grand Strand area of South Carolina north of Georgetown to Cherry Grove. This is presumably due to the absence of large estuaries. In spite of extensive menhaden research in North Carolina *C. carolinensis* has not yet been reported from that state. However, several individuals were recently recovered in Virginia from the collections of the Virginia Institute of Marine Science (VIMS). Three individuals were collected on 25 March 1959 from menhaden in the York River. Another individual was found on 21 January 1965 unattached to a host in a suspended tray of oysters at the VIMS pier, York River, Virginia (37°14′46″N; 76°30′02″W). In spite of extensive parasitological research by VIMS, it has not been found there since.

Like most truly estuarine invertebrates, *C. carolinensis* displays a remarkable tolerance to fresh water. During the study it was collected in water ranging from 32.5‰ at 10.5° C to 4.5‰ at 19.4° C, but most individuals were collected in water from 10 to 20‰. In the laboratory a series of salinity tolerance experiments was undertaken in which leeches were taken from water at 28‰ and placed immediately into salinities of 0, 1.62, 3.24, 6.48, 12.19 and 25.85‰ (Sawyer, in preparation). Each container held five adult leeches, and the experiment was undertaken at 10° C as well as at 21.5° C. At each salinity the leeches clearly had a greater tolerance to fresh water at the lower temperature than at the higher temperature. For example, they withstood pond water 12 days at 21.5° C and for 51 days at 10° C. At 10° C they lived indefinitely from 3.24 to 25.85‰, while at 21.5° C they lived indefinitely only in 25.85‰. When the leeches were slowly conditioned to pond water the results were similar.

Feeding

The infestation rate of *C. carolinensis* on menhaden in South Carolina estuaries is remarkably high. For example, at Beresford Creek on 23 February 1972, 67.7% of the 41 menhaden (85 to 138 mm, SL) collected were infested with 210 leeches. Most of the fish harbored from 2 to 17 leeches, but two fish (115 and 126 mm, SL) each had 36 leeches in their mouth cavities. On the whole it appears that the larger fish harbor more parasites. On 19 December 1972, 348 leeches (mostly from 4 to 10 mm in length) were found in the mouth cavity of one large menhaden (227 mm, SL). Similarly, on 8 February 1973 of 11 menhaden examined 10 (99–153 mm, SL) had from 4 to 75 leeches in their mouth cavities. Other samples of menhaden yielded similar results, some of the smaller samples occasionally yielding up to 100% infestation. Leeches were most commonly found on menhaden of 96 to 126 mm, SL, but some were found on fish of 91 mm to 277 mm, SL.

Throughout the three years of the study all sizes of *C. carolinensis* collected from the menhaden occurred exclusively in their mouth cavities, none being found on the body nor the fins. None of the leeches was actually seen sucking the blood of the fish, but the crops of most were engorged with blood. In the laboratory, however, the leeches were observed feeding on the clingfish, *Gobiesox strumosus*, the white catfish, *Ictalurus catus*, and the mullet *Mugil cephalus*. In each case feeding was on various regions of the body outside the mouth cavity and lasted for at least 6 to 8 hours. The body of the leech was sharply bent and the suckers were closely positioned. In some cases the fish reacted with a frenzied darting motion which occasionally dislodged the leeches.

Under natural conditions *C. carolinensis* feeds almost exclusively on clupeid fish, particularly on menhaden. In fact, the overwhelming majority of the many hundreds of leeches collected from fish in the Charleston area were taken from the Atlantic menhaden, *Brevoortia tyrannus*. The most notable exception was the occurrence on 21 April 1971 of numerous leeches in the mouth cavities of the Blueback herring, *Alosa aestivalis* (Mitchill), another clupeid fish with feeding habits similar to those of *B. tyrannus*. On 13–14 December 1972 small individuals were found in the mouth cavity of the blueback herring. No other *C. carolinensis* were collected on blueback herring, nor on other clupeid fish examined. However, one leech was found in the mouth cavity of the spot, *Leiostomus xanthurus,* and the summer flounder, *Paralichthys dentatus,* and one was collected from the ventral surface of the clearnose skate, *Raja eglanteria,* and the southern flounder, *Paralichthys lethostigma.* Considering that many hundreds of spots and dozens of flounders and rays were examined, it seems likely that these are just accidental relationships of no real significance.

The majority of blood-sucking glossiphoniid and piscicolid leeches leave their hosts and seek a solid substratum during the breeding season and *C. carolinensis* is no exception. In the late winter and early spring months, most of the engorged leeches leave the menhaden and become attached to oyster clumps and associated green seaweeds (*Ulva*) apparently the only suitable substrates commonly available to them in the mud-bottomed tidal creeks of South Carolina. At Beresford Creek on 8 February 1971 and again on 23 February 1972 and for weeks thereafter in each case, large numbers of leeches were dredged up with oyster clumps. It was also about this time, 13 March 1971 and 29 January 1972, that large numbers of engorged leeches were found on detached green seaweeds floating in the surf at Folly Beach.

The rate of digestion of ingested blood is remarkably slow and is evidently very efficient. During digestion the intestine was noticeably much darker than the crop. At 9° C the leeches lived for at least 10 weeks without further feeding, the dark fecal material slowly accumulating on the bottom of the aquaria. All the leeches in the laboratory eventually died.

Like most bloodsucking piscicolids examined, *C. carolinensis* is probably a vector for hemogregarines or hemoflagellates (see Becker and Katz, 1965a and 1965b; and Putz, 1972). Thus, *C. carolinensis* is a probable vector for the hemogregarine, *Haemogregarina brevoortiae*, recently described from menhaden from southern Florida (Saunders, 1964).

Behavior

Locomotion in *C. carolinensis* is in the "inchworm" manner typical of the Piscicolidae. The foresucker is extended until it attaches to the substratum; the hindsucker is then positioned near the midventral portion of the body and slides down the body until it is positioned immediately posterior to the foresucker. When an individual is disturbed, the hindsucker is released and the body coils up like a snake with the head always dorsal. It remains in this position for a number of seconds before it crawls or swims away.

True swimming is strongly developed and can be sustained for periods up to one minute or even longer under ideal conditions. This is done by means of rapid, undulating motions of the flattened body and hindsucker. Swimming can be evoked by jarring the container, by dislodging the hindsucker from the substratum or by suddenly passing a shadow overhead. When the ventral nerve cord is severed, the end posterior to the transection displays swimming motions, whereas the anterior end displays crawling motions. Similarly, when the brain is excised the animal displays swimming motions. Unlike most leeches, *C. carolinensis* does not ventilate its body by undulating the entire body while the hindsucker is attached to the substrate, a manner reminiscent of swimming in position.

In an undisturbed aquarium *C. carolinensis* displays the curious habit of swimming to the surface of the water until the foresucker attaches to the surface film. Almost immediately the hindsucker is then attached and the foresucker is released in such a manner that the animal hangs upside down from the surface film for long periods. They can even crawl upside down along the surface film in the typical manner. This suspending behavior is most common in the smaller individuals (3 to 7 mm); a number of large individuals suspended only once, in a closed container that had been at room temperature for about 24 hours. Individual leeches were collected in plankton nets dragged at the surface of the water on 9 March and 11 March 1971, 3 March 1972, 6 February and 12 February 1973, when the water was about 11 to 12° C.

C. carolinensis displays a strong reaction to a sudden decrease in light intensity. Normally an individual maintained under constant light conditions will quickly respond to a shadow passing overhead with random "searching" motions of the foresucker. In extreme cases the passing shadow will evoke crawling, coiling or even swimming. Individuals which have had their two pairs of eyes removed with a scalpel blade also display the typical "searching" motions in response to a shadow. Unlike many leeches, this species does not display "searching" motions when its container is sharply vibrated. An individual engorged with blood is relatively inactive and usually displays little reaction to a shadow. Leeches maintained in constant dark will barely respond to a sudden light.

Habituation of leeches to a shadow was investigated. Ten leeches were maintained in individual containers at room temperature. The light source was a microscope light positioned about 40 cm from the animals. Experiments were conducted by passing a shadow at intervals of five seconds after the animal became quiescent. The duration of the "searching" response was timed until five negative responses were observed. Stronger responses were observed with the shadow originating at the light source itself rather than originating immediately above the animal. During every experiment the duration of the response gradually decreased to the point that the leech did not respond at all. In the original series of experiments this point varied among individuals from 14 to 109 trials. In a second series of experiments one hour later this point varied from 13 to 25 trials and similar results were obtained in a third series, on a few individuals, 24 hours after the second series of experiments.

The duration of the longest response for each individual varied from 4.0 to 10.0 seconds in the first series of experiments and from 3.8 to 10.0 seconds in the second series. The longest response of each individual occurred in the second to tenth trial in the first series of experiments and in the first to sixth trial in the second series.

C. carolinensis, which has unusually large chromatophores, dramatically demonstrates physiological color change in response to changes in ambient light and temperature (Sawyer and Dierst-Davies, in press). The leeches darken in the light and blanche in the dark, and to a lesser extent darken under warm water temperatures (23.0° C) and blanche under cold water temperatures (9.0° C). The leeches do not adapt to background coloration. It was demonstrated by severing the ventral nerve cord that color response is probably under neurohumoral control from the brain.

Respiration is probably aided by pulsating vesicles which are arranged metamerically along the lateral margins of the body. At 27° C the vesicles pulsated 57 to 64 times per minute, and at 18.5° C 30 to 41 times per minute. However, there is a great deal of variation in the rate of pulsation and there does not appear to be much coordination between the vesicles. When the ventral nerve cord was severed the vesicles at both ends of the body continued to pulsate.

Reproduction and development

Reproduction in *C. carolinensis* involves some interesting aspects which are unique to the Piscicolidae. The anatomy of the unusual reproductive system is described in detail by Sawyer and Chamberlain (1972). Courtship and mating was observed on numerous occasions both in the laboratory and in nature. During courtship two individuals approach one another and make agitated searching motions over one another's body with their foresuckers. At the same time the male bursae of one or both individuals are protruded to form a conspicuous cylindrical penis, a rare structure in the Piscicolidae. From time to time the anterior ends of their bodies tightly intertwine to culminate in true copulation. Copulation in this species involves the introduction of the penis into the spacious bursa of the other individual. It is doubtful whether simultaneous reciprocal copulation takes place in these hermaphrodites. Unlike most other piscicolids that have been studied, two individuals remain *in coitu* for 24 hours or even longer. A series of cross-sections through two individuals fixed *in coitu* revealed that a spermatophore containing sperms is deposited into the unique seminal receptacle. Mating couples face in the same direction or end to end (Fig. 2A) and they frequently vary positions by moving the suckers. When disturbed, the mating individuals often attempt unsuccessfully to pull themselves apart. In fact, it is relatively difficult even to pull them apart manually. Two individuals which had each been tightly ligated with a silk thread as a part of another experiment mated successfully 3½ hours later.

Mating normally takes place within a day of collection and continues with decreasing frequency for many weeks. Mating is encouraged by a change in water, light or temperature conditions. In nature, a mating pair was collected on 29 April 1972 on a piece of floating seaweed. Interestingly, mating was never observed on menhaden but was observed under the mouth of a mullet under laboratory conditions.

Cocoon deposition was observed on numerous occasions in the laboratory (Figs. 2, D–G). The individual places the clitellar region of the body adjacent to the glass substratum for a few minutes before the whitish ring of clitellar secretions become evident (Fig. 2D). When the ventral portion of this sticky ring firmly adheres to the glass, the body rotates itself within the ring, presumably to free the ring from the body wall (Fig. 2E). After a minute or so of rotation, the body anterior to the ring elongates (Fig. 2F) tremendously in preparation for the swift posterior movement which pulls the body through the ring (Fig. 2G). Unlike many leeches, *C. carolinensis* gives no further care to the helmet-shaped (diameter, 0.5 mm) cocoons (Figs. 2, B–C) which harden and become dark

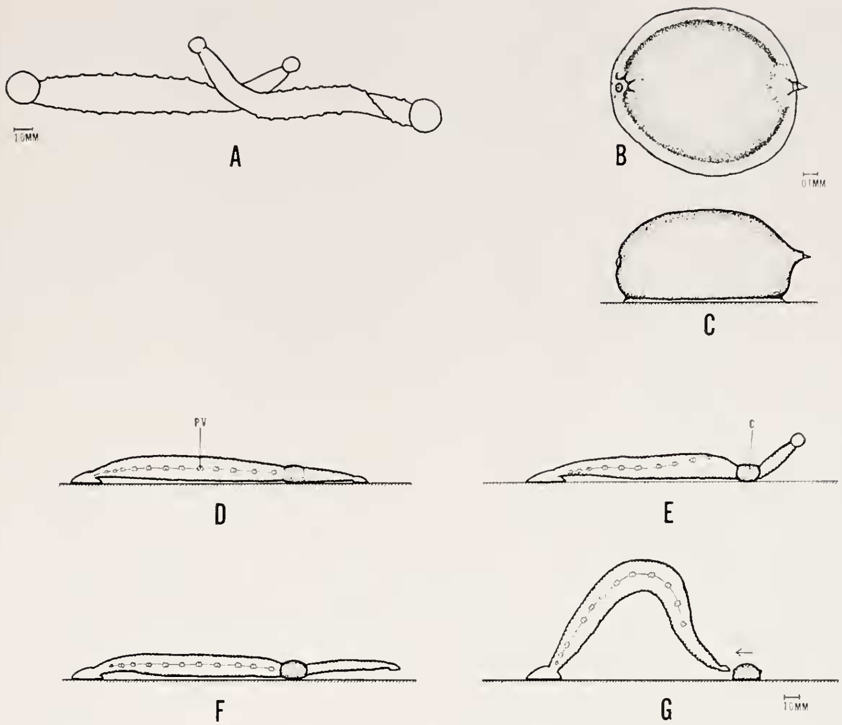

FIGURE 2. *Calliobdella carolinensis;* A, two copulating individuals; B, dorsal view of cocoon; C, lateral view of cocoon; D–G, cocoon deposition. See text for further explanation.

brown in about an hour. Like the cocoons of all other Piscicolidae, each cocoon contains only a single egg.

Throughout cocoon deposition, which takes 3 to 4 minutes, the hindsucker is firmly attached, whereas the foresucker is free to move about. The individual usually remains attached to the same spot and deposits many more cocoons. Often the deposition of cocoons is interrupted by mating, again without moving the hindsucker. Sometimes the individual moves to several other spots to deposit the rest of the cocoons.

In the laboratory the first cocoons were deposited on 12 February 1971 and 26 February 1972 by individuals collected on 8 February 1971 and 23 February 1972 when the water temperature was 9.2° and 9.4° C, respectively. Fourteen individuals collected on 8 February 1971 first mated and deposited cocoons on 12 February. Most of the cocoons were deposited between 15 and 19 February, and the last of a total 211 cocoons was deposited on 24 February. Most of the cocoons were aggregated into ten clumps of 7 to 29 cocoons. Assuming that 10 individuals were responsible for the aggregations, 21.1 cocoons per individual can be conservatively estimated. Similarly, two leeches collected on 5 March 1973 shortly afterwards deposited 41 cocoons.

In spite of repeated attempts, no cocoons were ever found in nature, but they will probably be found on the green seaweed *Ulva* or on oyster shells, the only common solid substrates suitable for cocoon deposition in the mud-bottomed tidal creeks. In the laboratory cocoons deposited on 14 February 1973 on oyster shells and on the walls of the aquarium hatched nine weeks later (water 21.5° C). In April and May 1971 and 1972, similar, but smaller, helmet-shaped cocoons of the swimming marine triclad, *Bdelloura rustica* Verrill, were frequently encountered on *Ulva* and at first confused with the cocoons of *C. carolinensis*. In laboratory containers maintained at 9° C the leech cocoons failed to hatch each year by May, at which time they were discarded. The leeches are about 3.0 mm long when they hatch from cocoons. The immature individuals (3 to 9 mm) differ only slightly from the mature individuals (9 to 29 mm) (Sawyer and Chamberlain, 1972). The former are more cylindrical with no distinction between the urosome and the trachelosome. Moreover, the body walls of the immature forms are usually translucent white, becoming more darkly pigmented with age. In the smallest individuals the lateral vesicles are small and internal, and the coelomic system is somewhat reduced. Almost identical differences between the immature and mature individuals were noted in the marine leech *Oceanobdella blennii* by Sawyer (1970).

Apart from the study by Sawyer (1970), the problem of growth in leeches has received very little attention. An analysis of growth patterns in *C. carolinensis* was undertaken on a population of 91 individuals ranging in size from 5 to 25 mm, collected on 13 March 1971 at Folly Beach. For each individual, which had been relaxed with 70% ETOH and fixed with formalin, the total length (L) of the body, the maximum width (W) of the body, and the maximum widths of the foresucker (F) and the hindsucker (H) were measured with an ocular micrometer. The data were analyzed for variance and non-linear regression with the aid of a digital computer. The analysis demonstrates clearly that, whereas some parts of the body grow at a faster rate than others, the growth of the four measured parts of *C. carolinensis* was essentially linear over the size range 5 to 25 mm (Table I).

Reconstruction of the life cycle of Calliobdella carolinensis

C. carolinensis normally lives in the mouth cavities of the Atlantic menhaden from mid-December to January. Starting in early January, when the water temperature decreases to about 12 to 13° C, the mature (9 mm and larger) leeches begin to leave the host in large numbers to reproduce in the tidal estuaries. Each individual deposits at least 20 cocoons in middle February when the water temperature is 9 to 10° C. Cocoons are probably deposited on oyster clumps or other solid substrates. Adults die by May, and no leeches are found until the following mid-December or early January when the young attach to juvenile (100 to 104 mm, SL) menhaden which have overwintered in estuaries. The newly emerged young are about 3.0 mm long and are good swimmers. Some probably find their hosts, which feed near the surface, by suspending themselves upside down on the surface film of the water. Others probably find their hosts by swimming toward a school of menhaden as it casts a shadow on the bottom.

Discussion

The epizootic outbreak of *C. carolinensis* reported above for January–March 1971 and 1972 is probably attributable to some factor(s) responsible for the in-

TABLE I

Regression analysis of growth in a population of 91 (one discarded) specimens of Calliobdella carolinensis *collected 13 March 1971 from Folly Beach. L, F, H and W mean total body length inclusive of both suckers, foresucker width, hindsucker width, and maximum body width, respectively. LW indicates an analysis of the relationship between the body total length and its maximum width,* etc.

	LW	LF	LH	WF	WH	FH
Regression coefficient	0.06718	0.03857	0.007440	0.39532	0.61057	1.54915
Standard error of regression coefficient	0.00623	0.00283	0.00582	0.03699	0.08932	0.12988
Correlation coefficient	0.75454	0.82346	0.80607	0.75150	0.58894	0.78605
Analysis of variance for simple linear regression (F value)	116.334	185.364	163.254	114.186	46.731	142.294
Sample size	90	90	90	90	90	90

creased numbers of survival of the offspring laid in January–March 1970 (or even one year prior). It now seems likely that an unusually cold spell and a high level of turbidity may have played an important role in this epidemic. As part of another study, surface water temperatures were recorded for every month from 1963 to 1972, at several routine sampling stations near Charleston. The coldest temperatures, which normally run 9.0–10.0° C, occur each year in January and February. Unfortunately, due to administrative changeovers no recordings were taken during the crucial period of January 1970. However, the records of the U. S. Weather Bureau at Charleston show that the monthly average air temperature of January 1970 (5° C) was the coldest for any month of any year since January 1940. This was well below the average January monthly air temperature of 10° C in the Charleston area. Unusually cold water temperatures were also recorded at all stations in February 1966 (6.8–7.2° C) and in January 1968 (3.5–4.8° C).

A related factor that may be overlooked is the high level of silt in the highly turbid (Secchi disk typically visible at 38 to 45 cm) waters of the estuaries of the Charleston area. This silt may have increased the turbidity of the water to such a level that, along with decreased water temperatures, natural predation on *C. carolinensis,* especially on the free-living breeding adults, was drastically reduced. The pollution ecology of leeches has been discussed in some detail elsewhere by Sawyer (1973) who presents experimental evidence supporting the supposition that natural predation by fish on leeches is decreased in turbid water. That external commensals and parasites are indicators of silt pollution receives circumstantial support from other species in this area. For example, the population levels of the brackish water leech, *Illinobdella moorei* Meyer, found on the white catfish, *Ictalurus catus* (Linnaeus); the marine leech, *Myzobdella lugubris* Leidy, found on the blue crab, *Callinectes sapidus* Rathbun; and the marine triclad, *Bdelloura rustica* Verrill, are much higher in this area than in similar regions of South Carolina where the water is much clearer.

Among freshwater leeches epizootic outbreaks are not unknown. Richardson (1928) presented a well-documented description of an outbreak of leeches (especially of *Helobdella stagnalis*) in the Middle Illinois River in 1925. In the region

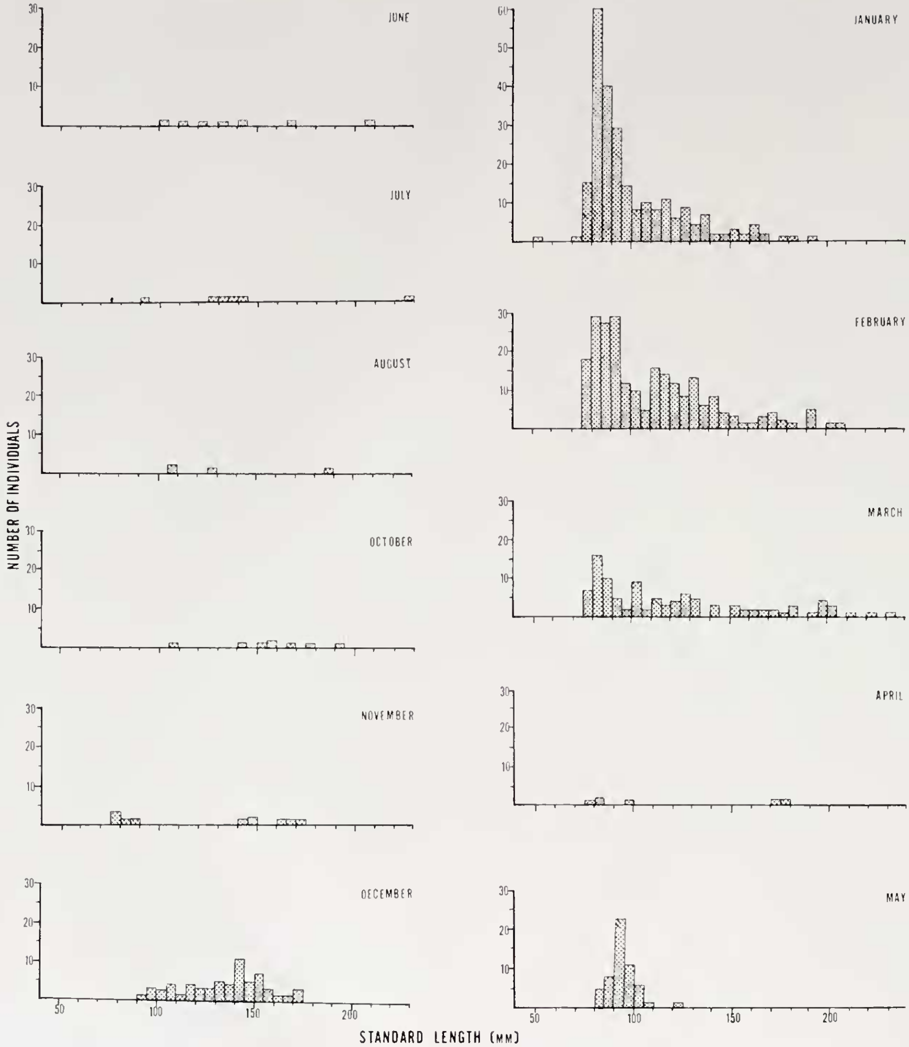

FIGURE 3. The relative abundance, seasonal occurrence and length frequency distribution (SL) of the Atlantic menhaden (*Brevoortia tyrannus*) collected by the South Carolina Marine Resources Center from Georgetown south to Hilton Head Island, S. C., for each month (excluding September) of 1970 and 1971 (combined data).

of the river most affected, the number increased from 1985 individuals per square meter in 1924 to as high as 29,107 in 1925. The total weight of the leeches changed from just over 200 kilograms per hectare (180 lb/acre) in 1924 to over 2800 kilograms per hectare (2550 lb/acre) in 1925. This meteoric rise in the concentration of leeches was undoubtedly due partially to the eutrophication of the river at that time. A similar outbreak of the fish leech *Piscicola punctata* was

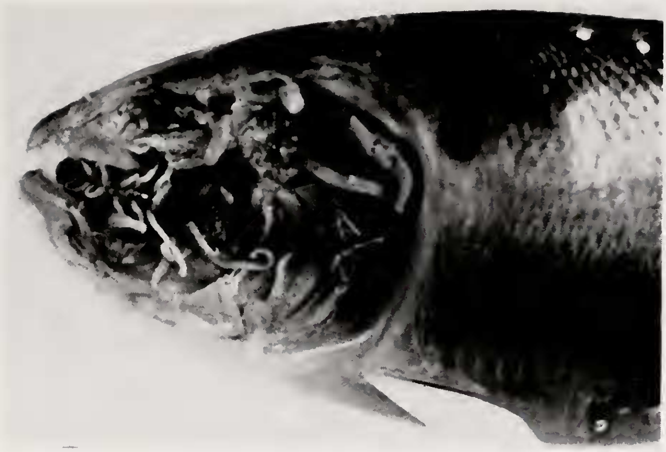

FIGURE 4. A specimen of *Brevoortia tyrannus* 155 mm, SL, collected near Charleston, S. C., 14 January 1971 displays 21 specimens of *C. carolinensis* and one isopod in the oral cavity.

described by Thompson (1927) in the nearby Rock River during the winter of 1925–1926. The course of this outbreak of *P. punctata,* a near relative of *C. carolinensis,* was remarkably similar to that described here for *C. carolinensis.*

One intriguing aspect of the life cycles of many piscicolid leeches is the conspicuous long absence of the leeches from the area soon after the adults leave the host to deposit their cocoons. For example, Halvorsen (1971) working with *Cystobranchus mammillatus,* Hoffman (1955) with *Cytobranchus respirans,* Thompson (1927) with *Piscicola punctata,* Becker and Katz (1965b) with *P. salmositica,* Halvorsen (1972) with *P. geometra,* Gibson and Tong (1969) with *Oceanobdella blennii,* and now this study with *C. carolinensis* all report intervals, usually from early summer to early winter, in which leeches are very scarce or are not to be found anywhere. The general consensus has been that the development or growth of the immature leeches is retarded during this period. Although our studies have shown that *C. carolinensis* can hatch in the laboratory after 9 weeks (water 21.5° C), no leeches were found in nature until the following winter. On 13–14 December 1972 (water 15.0 and 16.8° C, respectively) small individuals (6–10 mm) of *C. carolinensis* were found at Beresford Creek and elsewhere, in the mouth cavity of the blueback herring, *Alosa aestivalis,* as well as on menhaden. These individuals support the supposition that growth in this species is retarded until about mid-December.

Our investigations suggest that the life history of *C. carolinensis* is intimately associated with that of its host, the Atlantic menhaden. Unfortunately, very little is known about the ecology, population dynamics and life history of the menhaden in the numerous estuaries of South Carolina although extensive work in other portions of its range has been carried out by such investigators as June and Chamberlain (1958), June and Carlson (1971), McHugh, Oglesby and Pacheco (1959), Reintjes (1969), Wilkens and Lewis (1971) and Nicholson (1972). Briefly, these workers agree that the Atlantic menhaden ranges from central

Florida to New England. In the southern portion of its range, the adult menhaden spawns offshore during the winter. The larval fish (8 to 40 mm, TL) enter the estuaries after the yolk sac has been absorbed, moving up the estuaries to the freshwater transition zone, where they begin the transformation to juveniles. Becoming juveniles at about 113 mm (50 to 135 mm, SL), they form schools and enter all areas of the estuary, filter-feeding on plankton. They normally spend six to eight months in the estuaries before returning to the ocean in the autumn. A portion of the juvenile population will spend the winter in the nursery grounds, leaving the following year when they have attained approximately 179 mm, SL.

Based on data compiled by the South Carolina Marine Resources Center in routine monthly samples of menhaden caught in otter trawls in 1970 and 1971, the menhaden in the estuaries near Charleston have a definite seasonal occurrence (Fig. 3). They were most abundant in collections made in January, February and March. After February there was a monthly decline in the numbers caught in the trawls. The modes (and ranges) of the standard lengths of the individuals caught in the months of Febuary, April, June, September and December, 1970 and 1971 (combined data), were 90 (55–172), 94 (68–167), 113 (56–154), 115 (94–180) and 90 (62–145) mm, respectively. The abundance of menhaden in the winter months possibly reflects diminished net avoidance due to the low temperature of the water. *Calliobdella carolinensis* was especially prominent during this period. In our investigations *C. carolinensis* was found on menhaden ranging from 91 to 277 mm, SL, with most occurring on fish of 100 to 104 mm, SL (Fig. 4). Presumably these juveniles spent the winter in the estuaries where they became infected with the immature leeches.

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Summary

1. In January through March, 1971, and to a less extent the same period in 1972 and 1973, the marine leech, *Calliobdella carolinensis* Sawyer and Chamberlain 1972, was epizootic on the Atlantic menhaden, *Brevoortia tyrannus*, in the estuaries of South Carolina. Prior to this epizootic outbreak (epidemic) the leech was practically unknown.

2. The population dynamics of this outbreak were studied near Charleston, South Carolina, for three years, primarily in an estuarine creek and in the surf zone of an exposed beach.

3. *Calliobdella carolinensis* displays a seasonal occurrence which correlates with the temperature of the water. The leeches reach their greatest abundance in late February and early March, when the water is coldest (9 to 10° C). At that time the leeches breed and deposit at least 20 helmet-shaped cocoons. No leeches are found from the end of April until the following mid-December.

4. The leech occurs primarily in the estuaries of South Carolina, and is reported also from Virginia. It is not known south of South Carolina.

5. Feeding, reproductive behavior, growth, life cycle, salinity tolerance and other aspects of the biology of *C. carolinensis* are described.

LITERATURE CITED

Becker, B. D., and M. Katz, 1965a. Distribution, ecology and biology of the salmonid leech, *Piscicola salmositica* (Rhynchobdellae: Piscicolidae). *J. Fish. Res. Board Can.*, **22**(5): 1175–1195.

Becker, C. D., and M. Katz, 1965b. Transmission of the hemoflagellate, *Cryptobia salmositica* Katz, 1951, by a rhynchobdellid vector. *J. Parasitol.*, **51**: 95–99.

Gibson, R. N., and L. J. Tong, 1969. Observations on the biology of the marine leech *Oceanobdella blennii*. *J. Mar. Biol. Ass. U. K.*, **49**: 433–438.

Halvorsen, O., 1971. Studies of the helminth fauna of Norway, XIX. The seasonal cycle and microhabitat preference of the leech, *Cystobranchus mammillatus* (Malm 1863) parasitizing burbot, *Lota lota* (L.). *Norwegian J. Zool.*, **19**: 177–180.

Halvorsen, O., 1972. Studies of the helminth fauna of Norway. XXX. Seasonal cycles of fish parasites in the River Glomma. *Norwegian J. Zool.*, **20**: 9–18.

Hoffman, J., 1955. Quelque charactéres éthologiques de la Piscicolidée: *Cystobranchus respirans* Troschel. *Arch. Inst. Gr.-Duc. Luxemb. N.S.*, **22**: 223–225.

June, F. C., and J. L. Chamberlain, 1958. The role of the estuary in the life history and biology of Atlantic menhaden. *Proc. Gulf Carib. Fish. Inst.*, **11**: 41–45.

June, F. C., and F. T. Carlson, 1971. Food of young Atlantic menhaden, *Brevoortia tyrannus*, in relation to metamorphosis. *U. S. Fish Wildlife Serv. Fish. Bull.*, **68**(3): 493–512.

Llewellyn, L. C., 1965. Some aspects of the biology of the marine leech *Hemibdella soleae*. *Proc. Zool. Soc. London*, **145**(4): 509–528.

McHugh, J. L., R. T. Oglesby and A. L. Pacheco, 1959. Length, weight and age composition of the menhaden catch in Virginia waters. *Limnol. Oceanog.*, **4**(2): 145–162.

Nicholson, William R., 1972. Population structure and movements of Atlantic menhaden, *Brevoortia tyrannus*, as inferred from back-calculated length frequencies. *Chesapeake Sci.*, **13**(3): 161–174.

Putz, R. E., 1972. *Cryptobia cataractae* sp.n. (Kinetoplastida: Cryptobiidae), a haemoflagellate of some cyprinid fishes of West Virginia. *Proc. Helm. Soc. Washington*, **39**(1): 18–22.

Reintjes, J. W., 1969. Synopsis of biological data on the Atlantic menhaden (*Brevoortia tyrannus*). *U. S. Fish Wildlife Serv., Circ.* **320**: 1–30.

Richardson, R. E., 1928. The bottom fauna of the Middle Illinois River, 1913–25. Its distribution, abundance, valuation, and index value in the study of stream pollution. *Bull. Ill. Natur. Hist. Surv.*, **17**: 387–475.

Saunders, D. C., 1964. Blood parasites of marine fish of southwest Florida, including a new haemogregarine from the menhaden, *Brevoortia tyrannus* (Latrobe). *Trans. Amer. Microscop. Soc.*, **83**: 218–225.

Selensky, W. D., 1923. *Crangonobdella murmanica* n.g. n.sp., eine auf *Sclerocrangon* schmarotzende Ichthyobdellide. *Zool. Jahrb. Abt. Syst. Geo. Biol. Tiere*, **46**: 397–488.

Sawyer, R. T., 1970. The juvenile anatomy and post-hatching development of the marine leech, *Oceanobdella blennii* (Knight-Jones, 1940). *J. Natur. Hist.*, **4**: 175–188.

Sawyer, R. T., 1973. Pollution ecology of freshwater leeches. In press in C. W. Hart, Jr. and S. L. H. Fuller, Eds., *Pollution Ecology of Freshwater Organisms.* Academic Press, New York.

Sawyer, R. T., and N. A. Chamberlain, 1972. A new species of marine leech (Annelida: Hirudinea) parasitic on the Atlantic menhaden, *Brevoortia tyrannus. Biol. Bull.*, **142**: 470–479.

Sawyer, R. T., and K. Dierst-Davies, 1973. The physiology and phylogeny of colour change in freshwater and marine leeches (Annelida: Hirudinea). *Hydrobiologia,* in press.

Thompson, David H., 1927. An epidemic of leeches on fishes in Rock River. *Bull. Ill. State Natur. Hist. Surv.*, **17**: 195–201.

Wilkens, E. P. H., and R. M. Lewis, 1971. Abundance and distribution of young Atlantic menhaden, *Brevoortia tyrannus,* in the White Oak River Estuary, North Carolina. *U. S. Fish. Wildlife Serv. Fish. Bull.*, **69**(4): 783–789.